

AMENDMENTS TO THE CLAIMS

This listing of claims will replace all prior versions, and listings, of the claims in the application:

In the claims:

1-67. (Canceled)

68. (Currently Amended) A method for producing a cultured tissue construct, comprising,

(a) seeding human fibroblast cells capable of synthesizing an extracellular matrix on a porous membrane, wherein the membrane comprises pores that are about 3 microns or less in size, in a culture vessel in a chemically defined cell culture medium, wherein the chemically defined cell culture medium is free of undefined animal organ or tissue extracts and comprises ~~transferrin, ascorbate or an ascorbate derivative~~ the following:

(i) nutrient base medium;

(ii) insulin;

(iii) L-glutamine or an L-glutamine derivative; and

(iv) ascorbate or an ascorbate derivative;

(b) stimulating the fibroblast cells, which are at least at 80% confluence to synthesize, secrete and organize extracellular matrix components; and,

(c) continued culturing of the fibroblast cells ~~until the cells form a layer of synthesized to thereby produce a cultured tissue construct with an~~ extracellular matrix of at least about 30 microns thick, ~~with the and~~ cultured fibroblast cells contained within the synthesized extracellular matrix layer, wherein the extracellular matrix comprises:

(i) fibrillar collagen showing a packing organization of fibrils and fibril bundles exhibiting a quarter-staggered 67 nm banding pattern;

(ii) tenascin; and,

(iii) glycosaminoglycans;

and wherein said extracellular matrix is produced by the cultured fibroblast cells on one surface of the porous membrane ~~in the absence of exogenous extracellular matrix components during the culturing conditions, wherein the cultured tissue construct is capable of establishing a graft.~~

69-72. (Canceled)

73. (Previously Presented) The method of claim 68, wherein the human fibroblast cells are dermal cells.

74. (Previously Presented) The method of claim 68, wherein the ascorbate derivative is selected from the group consisting of sodium ascorbate, ascorbic acid and L-ascorbic acid phosphate magnesium salt n-hydrate.

75. (Currently Amended) ~~The product of~~ A cultured tissue construct produced using the method of claim 68, wherein the cultured tissue construct is free of undefined animal organ or tissue extracts.

76. (New) The method of claim 68, wherein the nutrient base medium is selected from the group consisting of Dulbecco's Modified Eagle's Medium (DMEM), Minimal Essential Medium (MEM), M199, RPMI 1640, Iscove's Modified Dulbecco's Medium (EDMEM), Ham's F-12, Ham's F-10, NCTC 109, and NCTC 135 nutrient base media.

77. (New) The method of claim 68, wherein the L-glutamine derivative is L-alanyl-L-glutamine.